

**Ecological Effects Branch  
Data Evaluation Report**

1. **Chemical:** Dithiopyr - Designated MON 7200 in study report.
2. **Test Substance:** Dithiopyr Technical, 92.2% ai  
Active Ingredient: Pyridinedicarbothiolic acid  
(2-difluoromethyl) -4-
3. **Study Type:** Twenty-one day chronic toxicity study using Daphnia magna under flowthrough conditions.
4. **Study Identification:**  
Study Laboratory: ABC Laboratories, Columbia, MD.  
Study Director: Blasberg, Jennifer  
Study Dates: March 2-23, 1992  
Study Identification: ABC Study #39666  
Sponsor: Monsanto Corporation  
EPA Identification: MRID 423333-01
5. **Reviewed by:** Brian Montague, Fisheries Biologist  
Ecological Effects Branch  
Environmental Fate and Effects Division  
(H7507C)  
7-2-92
6. **Approved by:** Les Touart, Section Supervisor  
Ecological Effects Branch  
Environmental Fate and Effects Division
7. **Conclusions:** The study as submitted is incomplete due to the lack of raw data to support the conclusions made by the study author. The study appears to have followed acceptable laboratory practices. Conclusions reached by the study author were that MATC limits based on the analysis of raw data not included in the study report were  $> 0.081 < 0.19$  mg/L. The 21 day  $EC_{50}$  was calculated to be 0.10 mg/L.
8. **Recommendations:** Registrant should submit raw data concerning individual daphnid weight, daily reproduction, and individual daphnid length to allow independent statistical analysis of these parameters as well as to confirm mean values presented in the report. This requirement is stated in the Agency's Standard Evaluation Procedures (page 6) for this type of study which are referenced in the study report itself (page 32).



9. **Submission Purpose:** Submitted to satisfy registration data requirements.
10. **Study Protocol and Design:** ASTM, USEPA, and OECD guidelines were used as guidance in preparation of study protocol.

**Test Organisms:** Daphnia magna were obtained from in-house cultures maintained at  $20 \pm 2^{\circ}\text{C}$  with 16D/8N photoperiod. Daphnids were fed algal suspension supplemented with trout chow. All organisms were first instar (<24 hours old) when selected for testing.

**Test Solution Preparation:** Diluter stock solutions were prepared on December 13 and 23 by addition of 0.2711 gms of test material to 250 ml of DMF solvent resulting in a 1000 mg/L diluter stock solution. No precipitate was reported and stock solutions were refrigerated after preparation. A primary stock solution of 1.02 mg/L was prepared from the diluter stock solution and nominal concentrations derived from this solution were 0.048, 0.096, 0.20, 0.40, and 0.80 mg/L for preliminary range study and 0.030, 0.060, 0.13, 0.25, and 0.50 mg/L for definitive testing.

**Study Design:** Four 1 Liter glass beakers were used for each treatment level, solvent control, and the controls. A Mount and Brungs design diluter system was used to supply the 28 test vessels (7 test groups). Ten daphnids per chamber (40 per treatment or control) were employed. The diluter system was designed to deliver 5.1 volume additions/vessel/24 hours. Diluter systems were started three days prior to study initiation. Survival, behavior, and abnormal effects were observed daily for parental and F1 generation daphnids. Offspring were counted and discarded every Monday, Wednesday, and Friday during the study. Temperature, D.O., and pH were measured on days 0, 4, 7, and 14 in alternate replicates. Water bath temperature was continuously monitored with a thermal data logger. After removal of the adults, young were collected by pouring dilution water over stainless steel screens and collecting the screened dilution water in 1L beakers. Dilution water was then returned to test beakers along with adults. At test termination the remaining adults were measured (helmet apex to base of posterior spine) and then dried for 48 hours at  $40^{\circ}\text{C}$  for mean daphnid dry weight determination. Time to first brood and young /adult/ per reproduction day were determined as measurements of reproductive success or impairment.

Statistical analysis procedures employed Dunnett's T Test, ANOVA, Dunnett one tailed multiple means comparison, and Shapiro Wilks normality testing procedures. Statistical significance used was  $p \leq 0.05$ .

11. **Reported Study Results:** In the preliminary range study 100% mortality was seen at 0.80 ppm nominal concentrations,

20% at the 0.40 mg/L nominal, and 0% mortality in 0.20, 0.096, 0.048, and control test chambers. All survivors produced offspring by day 8 of this portion of the study. The range study was terminated after eight days.

Based on the results of the initial study, 21-day definitive testing was begun with 40 daphnids per four replicate test level at nominal concentrations of 0.030, 0.060, 0.13, 0.25, and 0.50 mg/L. Measured concentrations were 0.027, 0.044, 0.081, 0.19, and 0.36 mg/L representing from 62% to 90% recovery. Later analysis of residues on the 0.36 mg/L test level beakers produced recoveries of 0.270 and 0.140 ug/L, thus indicating adherence of test material to the glass of the test vessel walls.

Control and vehicle control 21-day survival rates were identical (97.5%) and therefore pooled for analysis purposes. Both 0.19 and 0.36 mg/L test levels suffered 100% mortality of adults before day 21. Mean survival levels of the 0.027 mg/L, 0.044 mg/L, and 0.081 mg/L test levels were respectively 95%, 92.5%, and 75%.

Reported mean length for controls was  $4.466 \pm 0.12$  mm and for solvent controls  $4.43 \pm 0.11$  mm. These were judged equivalent by the laboratory and pooled in analysis procedures. Mean measured lengths for 0.027, 0.044, and 0.081 mg/L concentrations were  $4.44 \pm 0.08$  mm,  $4.39 \pm 0.011$  mm, and  $4.37 \pm 0.09$  mm, respectively. These were judged to have no significant variance from the controls. Mean dry weight of the controls and solvent controls were  $0.83 \pm 0.05$  mg and  $0.83 \pm 0.08$  mg. Mean weights for surviving treatment groups were  $0.84 \pm 0.03$  mg (27 ppb),  $0.81 \pm 0.06$  mg (44 ppb), and  $0.79 \pm 0.14$  mg (81 ppb).

All treatments and control populations showed times to first brood ranging from 6.3 to 7.0 days. No significant difference was felt to have occurred here although  $L^3$  treatment levels were delayed by an average of 0.5 to 0.7 days over controls.

Pooled young/adult/reproduction days were 10.4, 10.3, 10.6, 10.0, and 6.43 for the controls (pooled), 0.027, 0.044, 0.081 and 0.199 ppm test levels, respectively. Total average young were 1606, 1593, 1573, 14966, 1433, and 579 for controls, vehicle controls, 0.027, 0.044, 0.081 and 0.19 mg/L test levels. All surviving young were reported to appear normal.

Water quality parameters measured in three concentrations and the control on days 0, 4, 7, 14, and 21 appeared to remain within acceptable ranges during the study.

12. **Study Author's Conclusion:** "Based on statistical analyses of survival, adult mean length, adult mean weight, days to

first brood, and young/adult/reproduction day from this 21-day Daphnia magna chronic toxicity study, MATC limits were determined to be 0.19 (LOEC) and 0.081 (NOEC) mg/L. The geometric mean of the MATC limits is 0.12 mg/L. The 21-day EC<sub>50</sub> was calculated to be 0.10 mg/L. The day 21 EC<sub>50</sub> should be interpreted with caution since there was not more than a 7.5% partial mortality in any of the levels."

13. **Reviewer's Discussion:** Dithiopyr has been shown in laboratory studies to demonstrate extreme hydrolytic stability (1/2 life  $\geq$  1053 days). The study author has noted the detection of test material residues on the glass of beakers used as test vessels. Thus, the lower than would be expected recovery levels may be explained by the adherence of residues to the glass. Despite this factor, mean recovery levels were generally above 60% of the nominal estimated concentrations. Lower recoveries were generally in the higher concentrations. Solvent effects are not apparent as recovery was nearly 100% for the stock solutions. Study conditions appeared to remain relatively stable throughout the study. Though measurement of water quality parameters was performed on days 0, 4, 7, 14, and 21 only, the results indicate that pH, temperature, and D.O. remained relatively stable. Dissolved oxygen should be measured more often (at least twice/week). A complete analysis of adult length, weight, and young produced/day could not be performed by the Agency as the study report has presented only summary tables of mean values not the raw data used to derive them. Before final acceptance of this study can be made, raw data used to derive growth and reproduction mean values must be forwarded to the Agency for review. This raw data may be transcribed if necessary for legibility, but it must include daily records and investigator observations.

**Adequacy of Study:**

**Category:** Incomplete

**Rationale:** Raw data needed to support author's conclusions has not been provided with the study.

**Repairable:** Yes - with submission of requested raw data.